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Puccinia subepidermalis sp. nov. and new records of rust fungi from Fairy Meadows, Northern Pakistan

N. S. Afshan [1*], A. N. Khalid[2], S. H. Iqbal[2], A. R. Niazi[2] & A. Sultan[2]

[1]pakrust@gmail.com*
Centre for Undergraduate studies, University of the Punjab
*Quaid-e-Azam Campus, Lahore, 54590, Pakistan**

[2]Department of Botany, University of the Punjab
Quaid-e-Azam Campus, Lahore, 54590, Pakistan

Abstract — Five rust species from Fairy Meadows in the Northern Areas of Pakistan are herein described from a recent urediniological survey. *Puccinia subepidermalis* infecting *Carex curta* is proposed as a new rust species from Pakistan and is distinguished from similar *Puccinia* sp. found on the same host. *P. praegracilis* infecting *Agrostis stolonifera* is reported as a new record from Pakistan, and *P. nepalensis* infecting *Rumex nepalensis* and *P. hieracii* var. *hieracii* infecting *Picris nuristanica* are new reports from Fairy Meadows, Northern Areas of Pakistan. A re-description of *Puccinia violae* infecting *Viola caespitosa* including SEM photographs is presented.

Key words — Kaghan valley, Nanga Parbat, *Puccinia caricis*

Introduction

The Northern Areas of Pakistan, lying under the great mountain ranges of Himalaya–Karakorum–Hindu Kush–Hindu Raj and Pamir surrounded by high peaks of 6500–8600 meters, is the most spectacular and fascinating region of Pakistan. In the heart of northern Pakistan, Fairy Meadows is located at the base of Nanga Parbat, which, at 8126 m, is the 9th highest mountain in the world and second in Pakistan after K2. The Fairy Meadows are lush green alpine pastures situated in the middle of a pine forest at an altitude of 3306 m. The pine forests skirting Fairy Meadows are one of the virgin forests in the North of Pakistan and home to a number of species of wild flowers, birds, and wildlife (Singh et al. 2004: 190–191). The altitudinal range of the Fairy Meadows vegetation belt is defined as montane belt. Although the montane belt on Fairy Meadows/Nanga Parbat is by far the richest in species number and potential

* Corresponding author

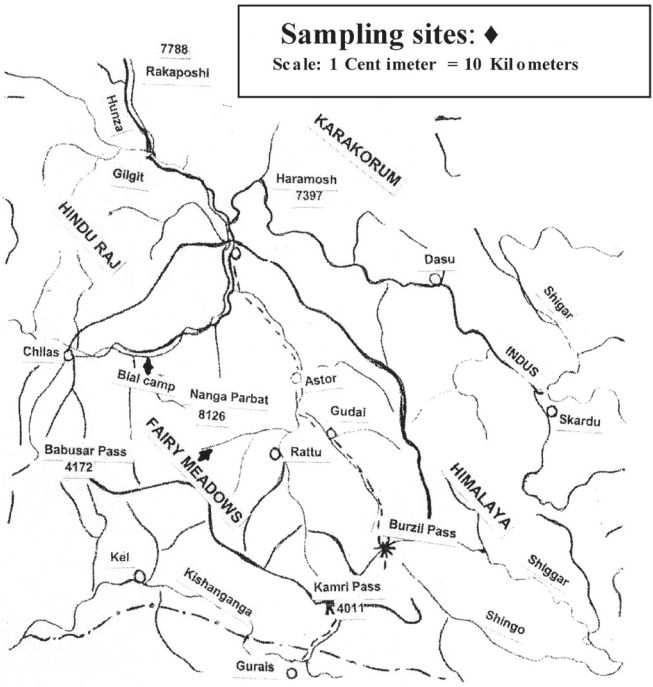


Fig. 1. Map of Fairy Meadows, Northern Areas of Pakistan, showing sampling sites

differentiation of vegetation types, it is floristically depauperate as compared to the outer Himalayan slopes (Troll 1939).

Locally, *Leontopodium campestre* Hand.-Mazz. and other herbs resistant to drought and grazing are plentiful. The ground layer of dry pure *Pinus wallichiana* A.B. Jacks. forests of the lower altitudes is widely dominated by a thin compost of dead pine needles. The companion flora of herbs and shrubs is relatively rich in species including *Androsace thomsonii* (Watt) Y. Nasir, *Artemisia japonica* Thunb., *Astragalus candolleanus* Boiss., *Bromus confinis* Nees ex Steud., *Geranium himalayense* Klotzsch, *Impatiens brachycentra* Kar. & Kir., *Rubus irritans* Focke, *Silene vulgaris* (Moench) Garcke and *Ribes nigrum* L. Moist areas by the stream-sides are characterized by *Salix* shrubberies (*S. wallichiana* Andersson, *S. sericocarpa* Andersson), occasionally with *Rosa macrophylla* Lindl., *Cotoneaster affinis* Hohen. ex Hook. f. and the herbaceous species *Rubus saxatilis* **L.** (Nüsser & Dickoré 2000: 13–26).

During a recent survey of the rust fungi in the Northern Areas of Pakistan, specifically Fairy Meadows, five species of rusts were encountered. Previously,

66 species of rust fungi have been reported from this area (Iqbal et al. 2009). In this paper, *Puccinia subepidermalis* on *Carex curta* is reported as new to science. In addition, *P. praegracilis* represents a new record for Pakistan while *P. nepalensis* and *P. hieracii* var. *hieracii* are reported for the first time from this region. *Puccinia violae,* previously reported from Pakistan, is re-described to illustrate important morphological features with the help of scanning electron microscopy.

Materials and methods

Specimens were collected from the Northern Areas (Fairy Meadows) of Pakistan (Fig. 1). Freehand sections of infected tissues and spores were mounted in lactophenol and gently heated to boiling. The preparations were observed under a NIKON YS 100 microscope and photographed with a digipro-Labomed and JSM5910 scanning electron microscope. Spores and paraphyses were drawn using a camera lucida (Ernst Leitz Wetzlar, Germany). Spores were measured with an ocular micrometer. At least 25 spores were measured for each spore state. The specimens were deposited in the Herbarium of the Botany Department, University of the Punjab, Lahore (LAH).

Enumeration of taxa

Puccinia subepidermalis Afshan, Khalid & S.H. Iqbal, **sp. nov.** Figs. A–B, Table 1

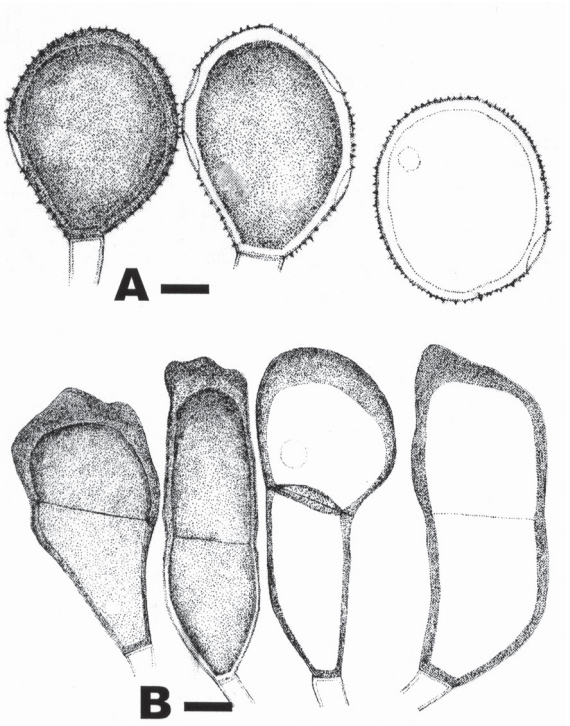
MycoBank MB 515049

Spermogonia et aecia ignota. Uredinia amphigena, subepidermalia, dilute brunnea. Urediniosporae globosae vel subglobosae, obovoideae; 23–27(–31) × 20–24 µm; membrana 1.5–2 µm crassa, pallidae-brunneae, echinulatae; poris germinationis 2–5, aequatorialibus, pedicellis hyalinis, usque ad 15 µm longis. Paraphysibus clavatis vel capitatis, hyalinae vel dilute flavidae, apice 18–22 µm crasso, membrana 2 µm crasso, usque ad 75–90 µm longo. Telia amphigena, subepidermalia, brunnea. Teliosporae 1–2 cellulares, clavatae, ellipsoideae vel oblongae; (34–)38–51(–58) × 15–20(–24) µm, membrana 2–3 µm crassa, apicaliter brunneae vel castaneo-brunneae, basaliter pallidae, pariete levi; apice truncato vel conico, 4–6 µm crasso; pedicello brunneo, usque ad 4–7× 8 µm.

Holotype: On *Carex curta* Gooden., with II and III stages, Pakistan, Northern Areas, Fairy Meadows, 3036 m a.s.l., 12th August, 2007. NSA #G35. (LAH Herbarium No. NSA 1094).

Etymology: Named due to the presence of subepidermal sori.

Spermogonia and aecia not found. Uredinia amphigenous, subepidermal, pale brown, 0.6–0.8 × 1.9–2.0 mm. Urediniospores globose to subglobose or obovoid, 23–27(–31) × 20–24 µm (mean 19.4 × 18.8 µm); wall 1.5–2 µm thick, orange to golden brown, echinulate; germ pores 2–5, equatorial; pedicel hyaline, 5–6 µm wide and up to 15 µm long. Paraphyses clavate to capitate, hyaline to pale yellow, head 18–22 µm wide, wall up to 2 µm thick, 75–90 µm long. Telia amphigenous, subepidermal, dark brown, 0.4–0.7 × 0.6–0.8 mm. Teliospores 1–2 celled, ellipsoid to oblong or clavate; wall 2–3 µm thick,



Figs. A–B: Lucida drawings of *Puccinia subepidermalis* (type) (A). Urediniospores (B). Teliospores. Scale bars = 10 μm.

golden brown to chestnut brown but paler basally, smooth; (34–)38–51(–58) × 15–20(–24) μm (mean 44.3 × 15.4 μm); apex truncate or conical to obliquely conical, 4–6 μm thick; germ pore 1 per cell, obscure; pedicel short, brown, 4–7 μm wide and up to 8 μm long.

Comments: Rust fungi on *Carex* spp. previously reported from Pakistan include *Puccinia caricina* DC. 1815 and *P. caricis-filicinae* Barclay 1889 on *Carex filicina* Nees, and *P. dioicae* Magnus 1877 and *P. pakistani* S. Ahmad 1961 on *Carex nubigena* D. Don (Ahmad et al. 1997). About seventy species of *Puccinia* have been reported on species of *Carex* throughout the world (Arthur & Cummins 1962, Wilson & Henderson 1966, Hiratsuka et al. 1992, Hennen et al. 2005).

Puccinia subepidermalis is characterized by the presence of clavate to capitate uredinial paraphyses with short pedicels of teliospores. The presence of 1–2-celled teliospores with truncate or conical to obliquely conical apices also distinguish this species from other species reported on *Cyperaceae*. (A comparison of *P. subepidermalis* with similar species is presented in Table 1).

Puccinia subepidermalis resembles *P. caricis* Rebent. 1804 in the size of urediniospores. However, *P. subepidermalis* has clavate to capitate uredinial paraphyses and shorter pedicels (8 µm vs. 45 µm) of teliospores than those of *P. caricis.* Moreover, presence of 1–2-celled teliospores with truncate or conical to obliquely conical apices differentiates *P. subepidermalis* from *P. caricis,* which has two-celled teliospores with rounded apices.

The new species has a few characters similar to *P. caricis-japonicae* Dietel 1906 including size and spore wall ornamentation. But these species are differentiated by the presence of clavate to capitate uredinial paraphyses and 1–2-celled teliospores in *P. subepidermalis.*

Table 1. Comparison of *Puccinia subepidermalis* with similar *Puccinia* spp.

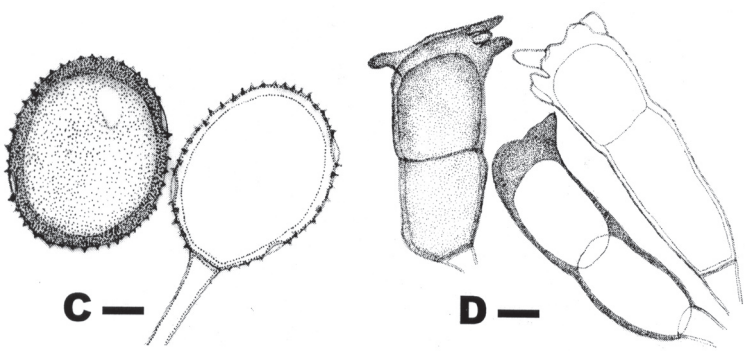
P. subepidermalis	*P. caricis*	*P. caricis-japonicae*	*P. caricis-shimidzensis*
Uredinia			
Amphigenous, subepidermal	Amphigenous, mostly hypophyllous, erumpent	Hypophyllous, subepidermal	Hypophyllous, subepidermal
Urediniospores			
(Sub)globose or obovoid	Subglobose or obovoid	Ellipsoid or obovate	Globose or ovate
Orange to golden brown	Light brown to brown	Yellowish brown	Yellowish brown
Echinulate	Echinulate	Minutely echinulate	Verrucose to echinulate
Germ pores (equatorial)			
2–5	3 (rarely 2 or 4)	3–4	2–3
Size (µm)			
23–27(–31) × 20–24	23–32 × 20–23	20–29 × 18–24	23–31 × 21–27
Paraphyses			
Abundant (clavate to capitate)	Absent	Absent	Absent
Teliospores			
Amphigenous, subepidermal, loculate	Hypophyllous, erumpent	Hypophyllous, naked	Hypophyllous, naked
1–2-celled	2-celled	2-celled	2-celled
Ellipsoid to oblong or clavate	Clavate	Clavate	Clavate or ellipsoid
Golden/chestnut brown, paler basally	Light brown	Yellowish brown to yellowish	Brown
(34–)38–51(–58) × 15–20(–24) µm	42–64 × 14–23 µm	39–56 × 11–18 µm	39–53 × 18–27 µm
Apex (µm thick)			
4–6	8–17	Unknown	9–10
Pedicel			
Brown, ≤ 8 µm long (4–7 µm diam)	Pale brown, 24–45 µm long	Hyaline or brown, 25 µm long	Hyaline or yellowish, 21 µm long

Puccinia subepidermalis is differentiated from *P. caricis-shimidzensis* Homma 1938 by spore size. *P. subepidermalis* has narrower urediniospores (20–24 µm vs. 21–27 µm) and teliospores (15–20(–24) µm vs. 18–27 µm) than *P. caricis-shimidzensis*. *P. subepidermalis* also has 1–2-celled teliospores with shorter (8 µm vs. 21 µm) pedicels and clavate to capitate uredinial paraphyses compared with the two-celled teliospores with longer pedicels of *P. caricis-shimidzensis*. The later also lacks uredinial paraphyses.

The new species also has few characters similar to *P. mandshurica* Miura 1928, but both species can be differentiated by the size of spores. *Puccinia subepidermalis* has larger urediniospores (23–27(–31) × 20–24 µm vs. 28–36 × 18–25 µm) and teliospores ((34–)38–51(–58) × 15–20(–24) µm vs. 36–50 × 10–15 µm) than in *P. mandshurica*. Moreover, the presence of echinulated urediniospores and 1–2-celled teliospores differentiates *P. subepidermalis* from *P. mandshurica* which has verrucose urediniospore wall ornamentation and two-celled teliospores.

Puccinia praegracilis Arthur, Hedwigia 37: 273 (1898) Figs. C–D

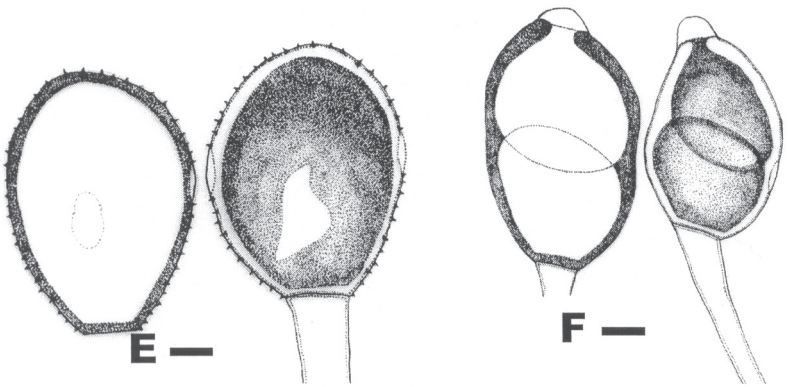
Spermogonia and aecia not found. Uredinia amphigenous, mostly on adaxial surface, yellow to yellowish brown. Urediniospores globose to ellipsoid or broadly obovoid, (16–)20–22 × (13–)16–21 µm; wall 1–1.5 µm thick, hyaline to pale yellow, echinulate; germ pores 5–7, scattered, obscure; pedicel colorless, minute. Telia subepidermal, mostly amphigenous or on abaxial surface, dark brown to blackish brown, with brownish stromatic paraphyses tending to divide sorus into locules, 0.2–0.5 × 0.3–0.8 mm. Teliospores elongated to obovoid or cylindrical to clavate, 37–44(–47) × 13–16(–21) µm (mean 41.6 × 16.9 µm); wall 0.5–1 µm thick at sides, 3–7 µm apically excluding digitations, golden brown to chestnut brown, smooth; with a few digitations, 3–6 µm long; pedicels short, mostly less than 15 µm long, 10–12 × 5–8 µm.



Figs. C–D: Lucida drawings of *Puccinia praegracilis* (C). Echinulate urediniospores (D). Teliospores. Scale bars = 10 µm.

Material examined: On *Agrostis stolonifera* L., with II & III stages, Pakistan, Northern Areas, Fairy Meadows, at 3,036 m a. s. l., 12th August, 2007, NSA #G 50. (LAH Herbarium No. NSA 1078).

Comments: *Puccinia praegracilis* has previously been reported on *Agrostis thurberiana* Hitchc. from Western Canada (Cummins 1971). It is a new record for Pakistan.



Figs. E–F: *Puccinia nepalensis*
(E). Echinulate urediniospores showing equatorial germ pores (F) Teliospores.
Scale bars = 10 µm.

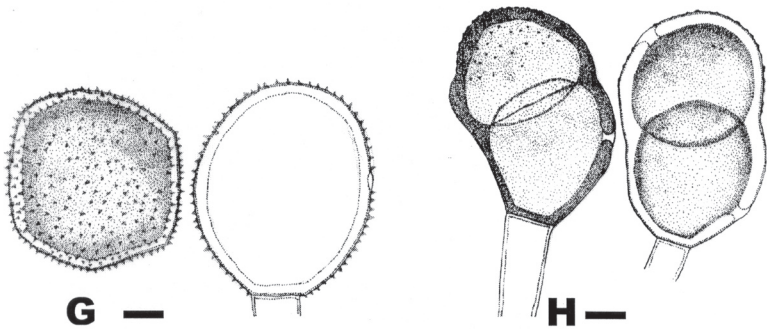
Puccinia nepalensis Barclay & Dietel, Hedwigia 29: 265 (1890) Figs. E–F

Spermogonia and aecia not seen. Uredinia intermixed with telia, amphigenous, scattered or in groups, sometimes circinate, cinnamon brown, pulverulent, 0.07–0.1 × 0.07–0.2 mm. Urediniospores globose or ovoid; wall 1–1.5 µm thick, echinulate, yellowish brown to cinnamon brown; 23–30 × 18–22 µm; germ pores 2, equatorial. Telia similar, dark brown to blackish brown. Teliospores ellipsoid, rounded at both ends, slightly constricted at septum, 29–38(–42) × 21–24 µm, with a hyaline papilla at apex next to septum, apex 4–5 µm thick; wall 1.5–2 µm thick, smooth, cinnamon brown; pedicel hyaline, 30 × 6–7 µm.

Material examined: On *Rumex nepalensis* Spreng., with II + III stages, Pakistan, Northern Areas, Fairy Meadows, at 3,036 m a.s.l., 12th August, 2007. NSA # G 17. (LAH Herbarium No. NSA 1072).

Comments: *Puccinia nepalensis* has previously been reported on *Rumex nepalensis* from Changla Gali, Hazara, Kaghan Valley, Murree, Muzaffarabad (Azad Jammu & Kashmir), Naran, Saiful Maluk and Swat (Ahmad et al. 1997).

Sultan (2005) reported *P. nepalensis* on *Rumex nepalensis* from Hunza Valley. This is a new record from Fairy Meadows.



Figs. G–H: *Puccinia hieracii* var. *hieracii*
(G). Urediniospores showing germ pore (H). Teliospores showing one germ pore in each cell.
Scale bar for G = 10 µm & H = 13 µm.

Puccinia hieracii (Röhl.) H. Mart., Prodr. Fl. Mosq. 2: 226 (1817)
var. ***hieracii*** Figs. G–H

Spermogonia and aecia not found. Uredinia amphigenous, mostly hypophyllous, surrounded by ruptured epidermis, brown, scattered over surface, early naked, 0.2–0.4 × 0.1–0.5 mm. Urediniospores globose to ellipsoid or sometimes angularly ellipsoid, 23–37 × 22–32µm (mean 30.4 × 26.6 µm); wall 2–3 µm thick, brown to chestnut brown, densely echinulate; germ pores 2–4, obscure, scattered; pedicel hyaline, minute, fragile, 12–15 × 6–8 µm. Telia mostly amphigenous, 0.2–0.6 × 0.2–0.4 mm, black. Teliospores ellipsoid to clavate, rounded at both ends, not or slightly constricted at septum, (29–)30–45(–48) × 21–30(–34) µm (mean 38.0 × 27.1 µm); wall 2–3 µm thick, verruculose, becoming smooth at lower side, chestnut brown; not thickened at apex; germ pore of upper cell sub-apical, of lower cell at equator or near septum; pedicel short, hyaline, 10–15 × 8–9 µm.

Material examined: On *Picris nuristanica* Bornm. (*P. hieracioides* L.), with II + III stages, Pakistan, Northern Areas, Fairy Meadows, at 3,036 m a. s. l., 12th August, 2007. NSA # G 12. (LAH Herbarium No. NSA 1059).

Comments: *Puccinia hieracii* var. *hieracii* has been reported on *Taraxacum officinale* Weber from Batakundi, Kaghan, Kalam, and Swat (Ahmad 1956a,b). It is a new record for Fairy Meadows, Northern Areas of Pakistan and *Picris nuristanica* is also a new host for this rust fungus from Pakistan.

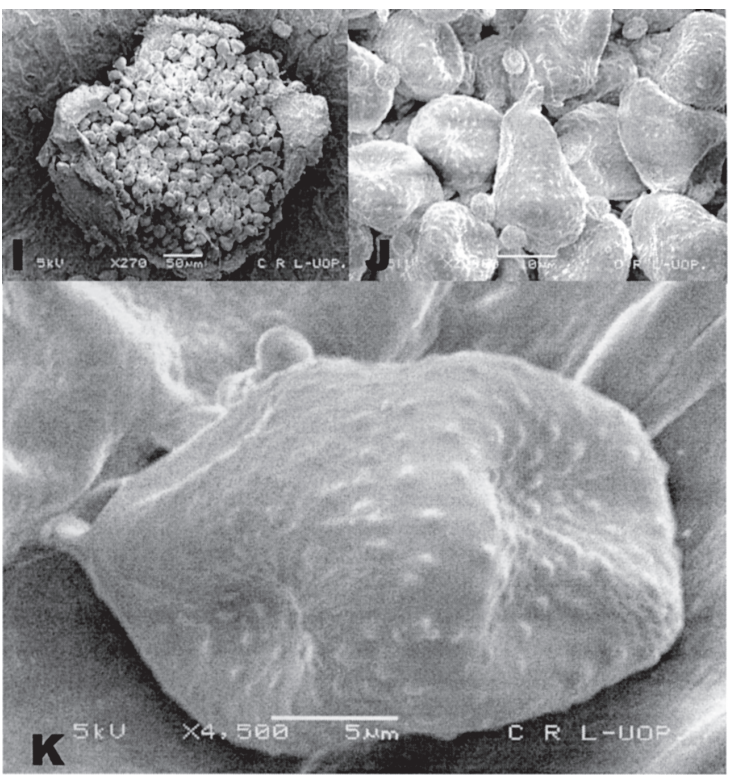
Puccinia violae (Schumach.) DC., Fl. franç., Edn 3, 6: 62 (1815) Figs. I–K

Spermogonia and aecia unknown. Uredinia hypophyllous, scattered or circinate, soon naked, pulverulent, brown, 0.1–0.2 × 0.2–0.4 mm. Urediniospores globose to obovoid or ellipsoid, 20–27 × 17–22 µm (mean 23.6 × 20.2 µm); wall 1.5–2 µm thick, pale brown to cinnamon brown, echinulate;

germ pores 1–2, equatorial; pedicel hyaline, short, 15–20 × 6–8 µm. Telia similar, dark brown to blackish brown, 0.08–0.1 × 0.2–0.8 mm. Teliospores ellipsoid to oblong, rounded at both ends or sometimes attenuated downwards, not or slightly constricted at septum, 25–32(–35) × 19–22 µm (mean 30.4 × 20.2 µm); wall 1.5–2 µm thick, smooth, chestnut brown; apex 4–5 µm thick, faintly verrucose with hyaline papilla over germ pores, pale in color; germ pore 1 per cell, pore of upper cell apical, of lower cell at septum, or both slightly subapical; pedicel short, hyaline, deciduous, 24–30 × 5–8 µm.

Material examined: On *Viola caespitosa* D. Don, with II + III stages, Pakistan, Northern Areas, Fairy Meadows, at 3,036 m a. s. l., 12th August, 2007. NSA # G 04. (LAH Herbarium No. NSA 1100).

Comments: *Puccinia violae* has been reported on *Viola biflora* L., *V. canescens* Wall., *V. caespitosa, V. indica* W. Becker, *V. rupestris* F.W. Schmidt, and *V. serpens* Wall. from Fairy Meadows, Hazara, Kaghan Valley, Shogran, and



Figs. I–K: *Puccinia violae* (I). SEM photograph of a telium containing teliospores (J). SEM photograph of teliospores (K). A teliospore showing verrucose wall ornamentation.

Swat State by Ahmad (1956a,b), Jørstad & Iqbal (1967), Ono & Kakishima (1992), Ono (1992), and Kakishima et al. (1993a,b).

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